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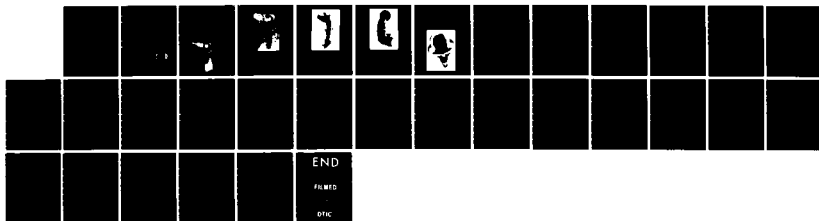
THE CRITICAL SIZE DEFECT AS AN EXPERIMENTAL MODEL FOR  
CRANIOMAXILLOFACIAL NONUNIONS(U) WALTER REED ARMY  
MEDICAL CENTER WASHINGTON DC J P SCHMITZ ET AL. 1985

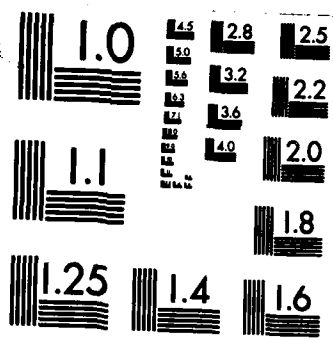
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The Critical Size Defect As An  
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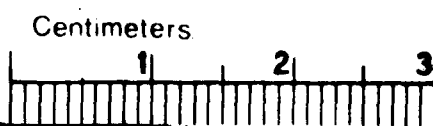
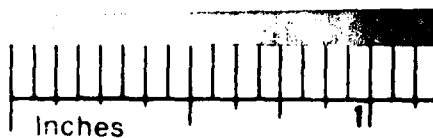
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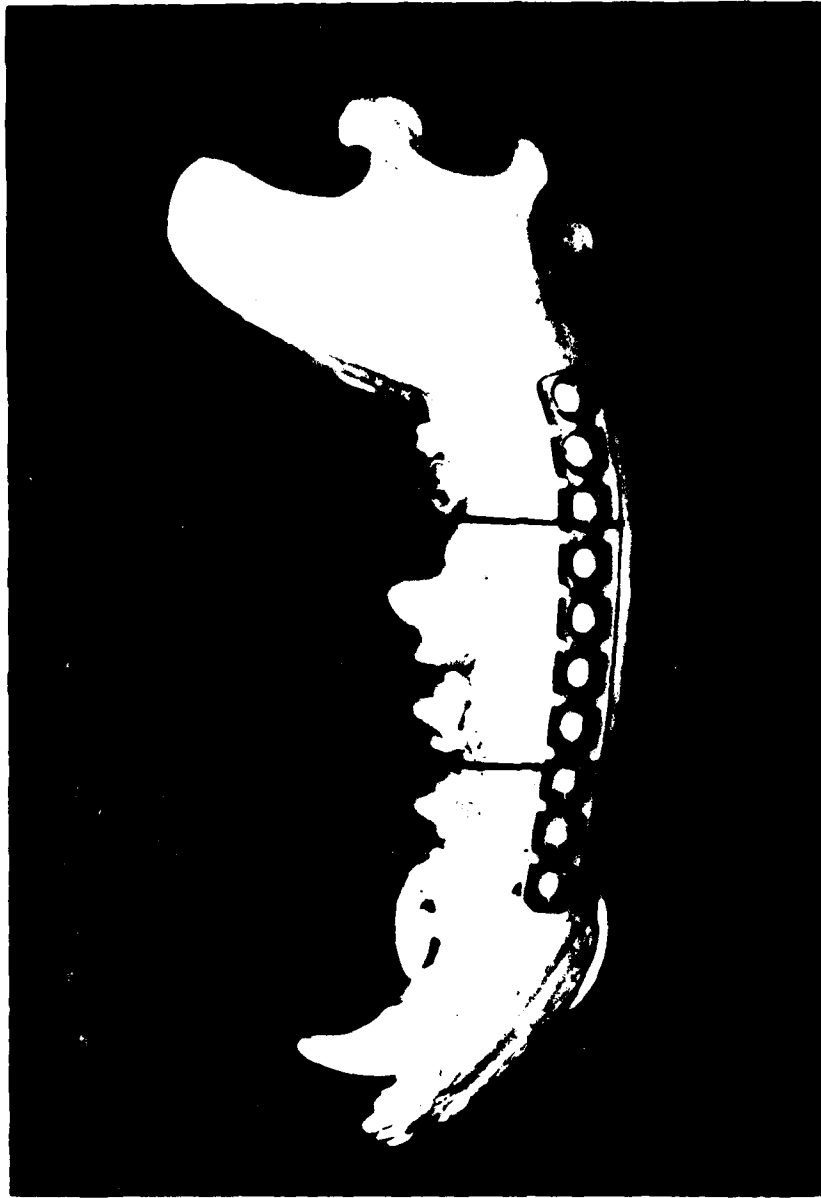
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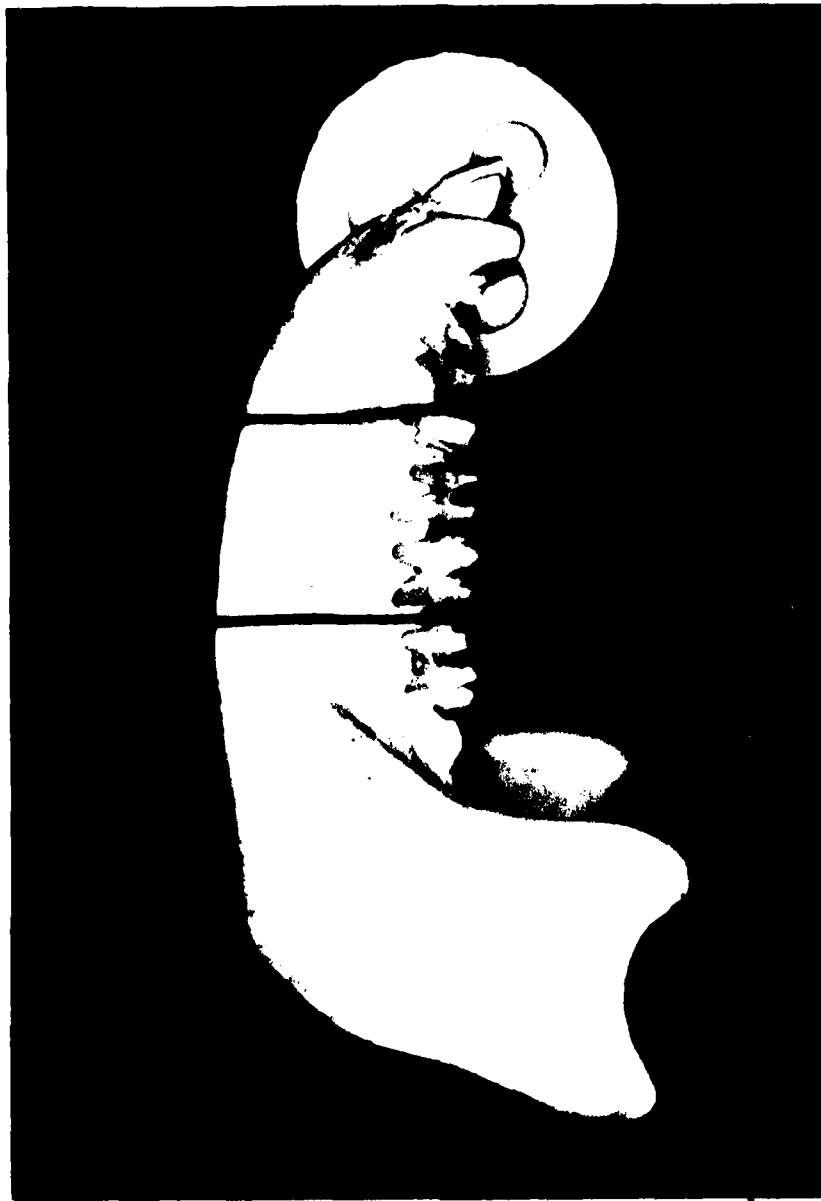
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## ABSTRACT

Little consistency has been maintained among research investigators in choosing an appropriate animal model for maxillofacial bone research. In an attempt to describe a protocol for the development of maxillofacial nonunions, experiments were reviewed which used calvarial and mandibular defects as models. The creation of experimental nonunions in the calvaria and mandible was found to be size-dependent. Defects of a size which will not heal during the lifetime of the animal may be termed critical size defects (CSD's). A rationale was postulated for testing bone repair materials (BRM's) using CSD's in a hierarchy of animal models. This protocol suggests that testing would be initiated in the calvaria of the rat and rabbit followed by testing in the mandibles of dogs and monkeys. While calvarial CSD's have been established in the rat, rabbit, and dog, research is still needed to describe the CSD in the calvaria of the monkey and the mandibles of dogs and monkeys.

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## INTRODUCTION

A variety of bone grafts and implants have found utility in oral and maxillofacial surgery. In reconstructive surgery, osteoinductive agents such as grafts and implants frequently are required either to repair maxillofacial defects or to augment the maxillofacial skeleton. Although novel bone grafts and implants have been evaluated in various species of experimental animals, little consistency has been maintained among investigators for the choice of an appropriate animal model.

Because the rate of bony repair varies inversely with order along the phylogenetic scale, experimental results obtained from animal wound models have been exceedingly difficult to compare.<sup>51</sup> Furthermore, the rate and amount of repair varies greatly among animals of the same species.<sup>15,22</sup> The animal models chosen to evaluate new bone repair materials often have consisted of immature, low-order phylogenetic species with a characteristically high potential for osteoneogenesis.<sup>2</sup> In animal models of this nature, the experimental wounds chosen as controls (those receiving no graft or implant) often have healed spontaneously. Immature animals of a given species can more actively repair an osseous defect than can an older one; therefore, a true test for a bone repair material should involve an adult animal.<sup>51</sup>

The amount of healing that will occur in a bony defect is in large part dependent upon wound size.<sup>26,56</sup> An experimental bony wound used to assess repair should, therefore, be large enough to preclude spontaneous healing. In this situation, the osteogenic potential of an implant or graft will not be considered unequivocal. An experimental bony wound of this nature may be termed a critical size defect (CSD). A CSD may be defined as the smallest size intraosseous wound in a particular bone and species of animal which will not heal of its own volition during the lifetime of the animal. Attempted

repair of a CSD results in the formation of fibrous connective tissue rather than bone.<sup>17,60</sup> Because there is no suitable animal model for the study of nonunions,<sup>56</sup> CSD's may be considered the prototype model for osseous nonunions and discontinuity defects. This paper reviews the literature on animal models and describes a protocol using CSD's for evaluating maxillofacial bone grafts and implants.

#### CALVARIAL DEFECTS

The bony vault of the cranium (calvaria) can be defined as that portion of the skull extending from the supraorbital ridge posteriorly to the external occipital protuberance.<sup>49</sup> It comprises the paired parietal bones, the squamous portion of the occipital and temporal bones, the squama frontalis, and a small section of the greater wing of the sphenoid.<sup>57</sup> The calvaria and the facial bones are pure membranous bones, with the mandible and the greater wing of the sphenoid being exceptions. Subtle differences exist between the microscopic and macroscopic structures and functions of the calvaria in different species; however, embryonic development is very similar.<sup>7,44</sup>

The biologic inertness of the skull compared to other bones can be attributed to a poor blood supply and a relative deficiency of bone marrow.<sup>51</sup> There is no nutrient artery in the human calvaria.<sup>57</sup> The middle meningeal artery provides the main cranial blood supply.<sup>11</sup> The calvarial bones receive accessory blood supply from arterioles emanating from the dural arteries to the inner table and to a lesser extent from the small arterioles of the outer surface.<sup>27,49</sup> Additionally, arterial blood enters the calvaria through the insertion of the temporalis muscle. However, because of the large area of the human skull which is devoid of muscle insertions, the blood supply to the human calvaria is poorer than in other mammals.<sup>57</sup> As a result, even

l defects in the human skull do not spontaneously repair.<sup>50</sup> In this  
ard, the regenerative capacity of the calvaria of experimental animals can  
onsidered better than that of humans.<sup>57</sup>

A calvarial wound model has many similarities to the maxillofacial  
on. Morphologically and embryologically, the calvaria develops from a  
rane precursor and thus resembles the membranous bones of the face.<sup>17</sup>  
omically, the calvaria consists of two cortical plates with regions of  
rvening cancellous bone similar to the mandible.<sup>17</sup> Physiologically, the  
icular nature of the cortical bone in the calvaria resembles an atrophic  
lible.<sup>2</sup>

Because the most severe test of a bone implant follows implantation in a  
l defect,<sup>50</sup> the calvaria has been a frequent site for the testing of bone  
ir materials. CSD's in the calvaria have been described in four animal  
ies.

#### RAT

Freeman and Turnbull<sup>19,61</sup> were the first to attempt the study of CSD's in  
calvaria. They showed that 2 mm diameter defects made through the  
osteum and parietal bone of 500 mg Wister albino rats failed to heal in  
ve weeks. Mulliken and Glowacki<sup>45</sup> and Glowacki et al.,<sup>21</sup> determined 4 mm  
eter defects to be the CSD in the parietal bone of 18-day-old Charles  
r rats. They reported that healing was unsuccessful at periods up to six  
hs, while evidence of bony repair was demonstrated at two weeks with  
neralized bone powder.

Tagaki and Urist<sup>60</sup> determined that 8 mm diameter defects created in the  
aria of six month old Sprague-Dawley rats were reduced to 5 mm in four  
s. No further healing of the defect was noted at twelve weeks. Healing  
he wounds commenced at the margins which became eburnated with the

formation of lamellar bone. The center of the defect healed by the formation of fibrous connective tissue. On the other hand, the experimental defects which were implanted with 10 mg of purified bone morphogenetic protein (BMP) developed chondroid at two weeks and were completely repaired in three weeks. This lends credence to the speculation that critical size defects can consistently repaired with osteoinductive agents.

As a general rule, weanling rats make unsatisfactory animal models for the evaluation of bone repair materials (BRM's) because of their tremendous ability to spontaneously repair large defects.

#### RABBIT

Kramer et al.<sup>35</sup> attempted to determine the CSD in the calvaria of six to ten pound New Zealand White rabbits. Evidence of healing in 8 mm diameter calvarial defects occurred at varying periods up to 16 weeks.

Frame<sup>17</sup> described the healing of 15 mm diameter defects in the calvaria of rabbits (crossbreed of New Zealand White and Half Lop). Using trephines, he created 5, 10, 15, and 20 mm diameter defects in the calvaria of 6.6 to 11.5 pound rabbits. At 24 and 36 weeks, the 15mm diameter defects had healed by the formation of fibrous connective tissue. Frame<sup>18</sup> subsequently tested an alloplastic material (calcium sulfate dihydrate/cyanoacrylate) in rabbits using 15 mm as the CSD. The control animals continued to retain a central, uncalcified area. The bone repair composite did not facilitate healing of the CSD and uncalcified areas were present at 24 and 36 weeks. Frame concluded that the material was not osteoinductive and was only moderately osteoconductive. His results furthered the thesis that only osteoinductive agents can consistently repair CSD's.

#### DOG

Efforts substantiating CSD's in the calvaria of mongrel dogs are well

nted. Friedenbergr and Lawrence<sup>20</sup> described the healing of 17 mm  
er defects in the calvaria of mongrel dogs. These wounds displayed less  
0% osseous repair at 20 weeks.

rolo et al.<sup>51</sup> used quantitative morphometry and determined that 20 mm  
er craniotomy defects in the parietal bone of mongrel dogs healed 20% at  
nths. When the defects were treated with bone sterilized with  
form/methanol and iodoacetic acid, an acceptable cranioplasty was  
ed 86% of the time.

rist<sup>62</sup> also has suggested 20 mm diameter craniotomy defects in dogs as  
D.

#### ONKEY

o data have been published establishing the calvarial CSD in any species  
lt monkey.

n creating experimental calvarial defects, a reproducible method should  
pted by the investigator for preparing standardized bony wounds.  
gular wounds are difficult to reproduce, therefore, circular trephines  
commended for creating experimental calvarial defects.<sup>1</sup>

#### ULAR DEFECTS

if all th al bones resected in oncologic surgery, the mandible is  
st frequently lved.<sup>5</sup> It is also the most frequent site in the  
ofacial region requiring bone grafts for the restoration of continuity  
nction.<sup>33</sup> Treatment of discontinuity defects that provides acceptable  
onal and esthetic results is extremely difficult because of the constant  
nt of the mandible in speech, mastication, and deglutition, and the  
ected contours which contribute to a person's self-image.<sup>6,12</sup> It is  
riate, therefore, that the determination of the maximum effectiveness of

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2) Allows a comparison of the rate of healing of a BRM vs. particulate cancellous marrow.

3) Permits the evaluation of a BRM in a site where it will eventually be used in humans (advantageous for future FDA approval of the material).



## Table II

suggested protocol for the testing of novel maxillofacial bone repair materials.

	Advantages
I. Begin testing in 8 mm diameter calvarial defects in rats (fig. 1).	1) Animals are inexpensive and may be procured in large numbers. 2) Only small quantities of the experimental agent are required for initial testing. 3) Particulate/gelatinous agents are well suited for implantation in this type of defect.
II. Continue testing in 15 mm diameter calvarial defects in adult rabbits (fig. 2).	This type of defect allows a solid implant to be evaluated in terms of its ability to be trimmed and morticed into a defect.
III. Finalize testing in discontinuity defects in the mandibles of adult mongrel dogs or non-human primates, or in non-human primate calvaria. (figs. 3,4, and 5)	1) Permits the evaluation of a BRM in a functional area (mandible).

of CSD's in various animal models.

	<u>Reference</u>	<u>Defect Size</u>	<u>Maximum Temporal Groups</u>
lvaria	Takagi and Urist <sup>60</sup>	8 mm	9-12 weeks
calvaria	Frame <sup>17,18</sup>	15 mm	24 weeks
lvaria	Prolo <sup>51</sup>	20 mm	24 weeks
ndible	-----	undetermined (greater than 20 mm?)	24 weeks
calvaria	Urist <sup>62</sup>	20 mm suggested	???
mandible	DeVore <sup>14</sup>	undetermined (suggested to be greater than 20 mm)	???

The concept of a CSD as a model for nonunions suggests a standardized set of controls for the evaluation of any potential maxillofacial bone repair material (BRM) (Table II). In this scheme, testing would be initiated first by evaluating the material in 8 mm CSD's in adult rats. This affords an inexpensive way to screen the BRM for toxicity and efficacy. Particulate or gelatinous-type materials are well suited for this type of evaluation. Once efficacy is established, the BRM should be tested using blocks or discs in 15 mm CSD's in rabbits. The thin bones of the calvaria in rats and rabbits are particularly well-suited for evaluation by high resolution radiography which allows correlation of radiographic with histologic findings. Additionally, the rabbit model allows for determining whether an implant can be trimmed or morticed into a circular defect. For mandibular-specific BRM's, evaluations should be in CSD's in dogs' mandibles because of the similarities to the human mandible in height, width, and length. Non-human primates are presently considered the benchmark animal model before proceeding to human-use studies. The final determination of the efficacy of a new BRM would be to compare its osteogenic potential to that of autogenous cancellous bone which is considered the grafting material of choice for maxillofacial defects.<sup>6</sup>

The concept of CSD's as a standardized set of controls using a hierarchy of animal models is a logical sequence for evaluating novel maxillofacial BRM's before Phase I human-use studies are contemplated. The protocol presented is considered to be a convenient series of animal model systems for testing BRM's.

possible only in the ramus area of small animals.

An ideal animal model for the study of mandibular discontinuity defects is available in the adult dog and monkey. Past research that attempted to define the CSD in the mandibles of mongrel dogs has shown that the CSD for adult mongrel dogs is probably between 20 mm and 40 mm, with 40 mm being the maximum size defect that can conveniently be created.<sup>10,28,29</sup> Although this ablation results in interruption of the arterial blood flow to the mandible, a retrograde blood supply subsequently develops providing adequate collateral circulation to the mandibular body.<sup>9,25</sup>

The key to success in avoiding problems of infection associated with ablative mandibular defects in dogs and monkeys involves effective isolation of the oral cavity from the experimental defect. Prophylactic antibiotics (penicillin or cefoxitin) administered at the time of surgery and post-operatively for five days is advocated. Osteomyelitis is rarely seen if the teeth in the experimental area are removed at least three to four weeks before creating the defect. This insures a healthy mucosal blood supply and a watertight mucosal seal. Additionally, a surgical approach through the submandibular region helps to avoid intraoral perforations. When preparing mandibular discontinuities, Penrose drains may be inserted to control seromas and hematomas.

Clinical efforts suggest that rigid internal fixation is the method of choice for managing mandibular nonunions.<sup>36,55</sup> Insertion of reconstruction plates according to AO/ASIF principles using non-self-tapping screws will ensure stability of the mandibular segments.<sup>58</sup> The tension-free adaptation of the periosteum over the implant also militates against mucosal dehiscence. If a dehiscence develops into the oral cavity, it is often amenable to daily oral hygiene therapy and saline irrigation.

attempt at bony repair. A nonunion will develop when the rate of fibrogenesis exceeds the rate of osteogenesis.<sup>63</sup>

The focal point for controlling the rate of fibrogenesis is the size of the defect in the bone. Key<sup>34</sup> observed that defects in long bones equal to one and one-half the diameter of the diaphyses routinely produced a nonunion. Heiple et al.<sup>23</sup> used an extraperiosteal resection equal to two times the diaphyseal diameter to produce a nonunion. Methods have been developed to consistently produce nonunions in the calvaria of rats, rabbits, and dogs using CSD's, however, no technique has consistently demonstrated nonunions in the calvaria of monkeys or in the mandibles of dogs and monkeys.

The quality of bony repair under optimum experimental conditions (non-infected defects) is markedly influenced by five experimental variables: 1) animal species,<sup>51</sup> 2) animal age,<sup>23,51,52</sup> 3) anatomic location of the experimental defect,<sup>22,46</sup> 4) size of the defect,<sup>26,53,56</sup> and 5) intactness of the periosteum.<sup>24,52</sup> While most of these factors are given strict attention in the design of an experiment, the issue of adult vs. young animals is frequently elusive. Most investigators tend to assess animal age using standardized weight charts, a practice fraught with inaccuracies. A more reliable technique for confirming skeletal maturity is radiographic evidence of closure of the epiphyseal plate of a long bone. Research is still needed to correlate animal weight with epiphyseal plate closure in various species of experimental animals.

Convenient animal models for the evaluation of calvarial defects in the rat, rabbit, and dog appear well established (Table I). However, the creation of discontinuity defects in the mandibles of small animals (mouse, rat, rabbit and guinea pig) is prohibitively difficult because of limited surgical access. As a result, through-and-through defects of the mandible may be

DeVore<sup>13</sup> has recommended a three-tooth defect (approximately 2 cm) as the CSD in the mandible of Cynomolgus monkeys with temporal groups extending to 26 weeks.

#### DISCUSSION

Although the bony nonunion is a frequent complication in the practice of orthopedics, it has not been easy to establish experimentally in animals.<sup>48</sup> Two techniques have been advocated for the creation of nonunions in animal models. One causes an artificially-created nonunion by impairing or preventing osseous regeneration. The second method results in a nonunion because the ablated segment is too large to be repaired by inherent osseous processes. This may be termed a CSD-created nonunion.

The techniques which impair normal union were reviewed by Neto and Volpon<sup>48</sup> and include: 1) maintaining movement at the osteotomy site via unstable fixation, 2) manipulating and distracting the fragments, 3) using no fixation at all, 4) using unstable osteosynthesis followed by manipulation, and 5) placing a foreign material in the defect to impair healing.

Instability, interposition of soft tissue, infection, inadequate blood supply, and nutritional and metabolic alterations have all been implicated as contributing factors in predisposing to nonunions in mandibular fractures and maxillofacial trauma.<sup>12,42</sup> However, in discontinuity defects, nonunions represents a space to be crossed and bridged by the normal processes of bony repair. The techniques associated with artificially-created nonunions, encourage fibrogenesis while discouraging osteogenesis through distraction, movement, and foreign bodies. CSD-created nonunions do not incorporate techniques associated with nonunions in fractures but encourage fibrogenesis while permitting any and all attempts at osteogenic repair by the proximal and distal fragments. The CSD-created nonunions thus represent a more physiologic

two-year-old Beagle dogs as the CSD, while Maughan<sup>43</sup> has suggested that 27 mm to 35 mm discontinuity defects in the mandibles of mongrel dogs heal by fibrous union at six months.

#### MONKEY

Roynes<sup>3</sup> created discontinuity defects in the mandibles of eight, six-year-old male Rhesus monkeys. Through a submandibular approach approximately 4 cm (from canine to third molar) of the mandible was excised unilaterally. After four weeks of healing, the animals were grafted with particulate cancellous marrow and fixated with a Sherman bone plate and a cage-type implant. Although no controls were used, osseous union occurred at six weeks with new cortical bone being present at six months. In the same experiment, Roynes also excised the mandible from second molar to second molar in eight adult male Rhesus monkeys. The area was grafted with particulate cancellous marrow and fixated with a chrome-cobalt implant. Clinical bony union at the symphysis was present at six months demonstrating the osteoinductive potential of particulate cancellous marrow.

Roynes<sup>4</sup> surgically removed the symphyseal area between the first mandibular molars of 14 Rhesus monkeys through an extraoral approach. Frozen allografts filled with autogenous marrow and cancellous bone were placed in eight animals and fixated with stainless steel orthopedic plates. Six animals received a metallic mesh crib filled with a composite of autogenous marrow and surface-decalcified bone. No controls were used in this study. Intraoral dehiscences and necrosis of the implants occurred two to four weeks postgrafting in nine out of fourteen animals. All the allograft composites showed varying amounts of nonvital bone matrix at six months, while four of the surface-decalcified allografts demonstrated regeneration of the mandible at twelve and twenty-eight weeks.

grafts. No control defects were utilized.

Holmes<sup>28</sup> prepared 20 mm ablative defects unilaterally in the mandibles of twelve adult mongrel dogs (15 to 20 kg body weight). Teeth in the area of the defect were extracted four weeks prior to creation of the defect. The mandible was surgically approached through a submandibular incision with fixation being achieved with a cast metal mesh tray and self-tapping screws (eight per tray). The defects in the experimental animals were restored with a coralline hydroxyapatite implant. The two control animals completely regenerated bone in the 20 mm defect in six months. Although all experimental defects had healed at six months, the absence of a well-documented CSD in the mandible of the adult mongrel dog makes the experimental results questionable.

Narang et al.<sup>47</sup> created five rhomboid-shaped defects (6 mm X 5 mm X 3 mm) in the body of the mandibles of five adult male mongrel dogs weighing 14 to 15 kg each. Evaluation of the defects at four, six, and eight weeks revealed 2 mm of bony regeneration in the central part of the defect. A rhomboid-shaped defect is not considered to be a standard defect and would be extremely difficult to reproduce accurately in experimental animals. This technique is not recommended for use as a standardized defect in animal models.

Stanley and Rice<sup>59</sup> created 1 cm X 3 cm unilateral defects in the inferior border of six mongrel dogs. Teeth had been extracted three weeks prior to preparation of the osseous defects. The oral cavity was not entered and preservation of the exterior border obviated the need of rigid fixation. This model was used to evaluate autoclaved, reimplanted autogenous bone. Although no control animals were used, microscopic evidence of bony union was seen at eight weeks.

Recently, Forbes<sup>16</sup> has suggested 17 mm x 6 mm defects in the mandible of



three months, the margin between host bone and graft was indistinguishable radiographically, while cortical bone was observed radiographically at six months. Although all animals received one million units of Bicillin® preoperatively and for five days postoperatively, two infections and one seroma developed and this required removal of the trays.

Leake and Habal<sup>37</sup> created 3 cm unilateral defects in the mandibles of mongrel dogs. Four weeks after extraction of the teeth, an extraoral approach was used to place a prefabricated alloplastic tray and particulate cancellous marrow graft. New bone capable of withstanding masticatory stresses was present at twelve weeks, postsurgically. Cortical bone was demonstrated by six months. Although no control animals were used in this study, the healing time of particulate cancellous marrow in dogs may be considered a standard against which other osteoinductive agents may now be compared.

Marciani et al.<sup>40,41</sup> studied 4 cm unilateral discontinuity defects in eight dogs and two monkeys. All animals received extractions followed in seven weeks by radiation therapy to the edentulous area. The dogs and monkeys subsequently were treated with iliac crest grafts and titanium mesh trays wired to the proximal and distal fragments; no intermaxillary fixation was used. All dogs exhibited healing in six months while the monkeys showed similar results at one year post-surgery.

Cummings<sup>10</sup> investigated 4 cm unilateral discontinuity defects in the mandibles of mongrel dogs. Experimental animals were treated with frozen mandibular bone implanted through an extraoral approach with fixation achieved by means of an extraoral biphase fixation appliance. Complete osseous healing of the experimental defects was observed at varying periods up to six months. Even though intraoperative and postoperative penicillin was used, focal areas of osteomyelitis were observed in the medullary portion of all the

Hjorting-Hansen and Andreassen<sup>26</sup> created 5mm, 6mm, and 8 mm circular defects bilaterally in the mandibles of six adult mongrel dogs. All the defects were prepared with a trephine in an edentulous area superior to the inferior alveolar canal. Defects on the left side were prepared through the buccal cortex only, while those on the right side were prepared through both cortices. At 16 weeks, the 8 mm defects demonstrated no regeneration of cortical plate while the 5 mm and 6 mm defects showed regeneration of at least the lingual cortical plate. All the 8 mm defects exhibited healing with fibrous tissue either extending from the buccal to the lingual mucosa or extending only from the buccal mucosa to the central areas of the mandible. The authors concluded that bony regeneration in the mandible was almost entirely dependent upon apposition from the lateral walls of the cavity or from the endosteum with minor contributions from the periosteum. They further concluded that three factors influenced bony regeneration: 1) the size of the defect, 2) the removal of one or both cortical plates, and 3) the elevation of periosteum.

Leake and Rappoport<sup>38</sup> evaluated the inductive capacity of iliac crest grafts in alloplastic trays placed in the mandibles of twelve adult mongrel dogs (10 to 12 kg body weight). Teeth were extracted unilaterally in the mandible and maxilla three weeks prior to the placement of the grafts and trays. Control defects 3 cm long were prepared through an extraoral incision in two animals; one received no fixation or tray, while the other received an empty tray wired in place. At six months, only a fingerlike projection of bone (10 mm X 3 mm) had formed at the superior border of the defect. There was no bony union between the proximal and distal fragments. The ten experimental animals received iliac crest grafts maintained in alloplastic trays. The grafts and trays were inserted through an extraoral approach. At

covered with Teflon® mantle-leaf to inhibit the formation of fibrous connective tissue. Defects in the right side of the mandible received no treatment (controls). At 52 weeks, the wounds covered with the mantle-leaf showed almost complete osseous regeneration whereas the two-wall defects demonstrated incomplete healing.

The CSD's used by DeVore and Kahnberg were not standardized and the model could have been improved by using a trephine to prepare the defects.

#### DOG

Calhoun et al.<sup>8</sup> were the first to attempt a well-controlled study of CSD's in the mandibles of mongrel dogs. Defects were prepared by the unilateral removal of the fourth premolar and its associated bone (approximately 15 mm). Intraoral and extraoral approaches to the mandible were used while fixation was achieved with either an extraoral stainless-steel splint or an internal fixation plate. Clinical, radiographic, and microscopic results from nine out of nineteen defects demonstrated evidence of bony union at 60, 90, and 120 days. Additionally, the animals did not tolerate the extraoral fixation device and it was frequently broken against the side of the cage requiring frequent replacement.

Huebsch and Kennedy<sup>29</sup> undertook a study to determine the CSD in the mandibles of six mongrel dogs. Teeth in the experimental area were extracted prior to creation of the defect and one week later the mandible was exposed by a submandibular approach. Ablative defects of 3 mm, 6 mm, and 9 mm healed uneventfully, with complete union evident at two months. The wider defects of 12 mm, 15 mm, and 18 mm in length exhibited bony union in four months but exhibited drainage either intraorally or extraorally up to the time of sacrifice. Penrose drains placed in the submandibular incision by the investigators may have alleviated this problem.

any graft material in maxillofacial surgery should be based on the ability of that graft material to reconstruct the mandible.<sup>6</sup>

Although the mandible develops through a process of branchiogenic (mixed) bone formation, physiological factors and not embryonic origin play the most important role in healing. In this regard, the mandible heals and remodels in a pattern similar to that of the tibia.<sup>46</sup> This similarity in healing is consistent with the functional forces acting on the bones: muscle-pull tension and body-weight compression in the tibia and muscle-pull tension and masticatory compression in the mandible. With the exception of the dog mandible, minimal attempts at documenting CSD's in the mandibles of other species are described in the literature. CSD's will be described in four animal species.

#### RAT

Kaban and Glowacki<sup>31</sup> and Kaban et al.<sup>32</sup> created 4 mm diameter through-and-through defects in the mandibular ramus of three month old Charles River rats. These defects failed to heal at 16 and 24 weeks. Demineralized bone powder was found to cause healing in these sites in two weeks.

#### RABBIT

DeVore<sup>13</sup> prepared osseous defects (approximately 5 mm long) through the inferior mandibular border of rabbits. Experimental wounds were deep enough to transect the inferior alveolar nerve and artery as well as the apices of the teeth. Although these defects completely healed at one year, a notch remained at the inferior border.

Kahnberg<sup>32</sup> studied the healing of 5 mm wide defects in the mandible of New Zealand White rabbits. The mandible was approached through a submandibular incision and two- and three-wall defects were subsequently created in pairs, bilaterally. Defects in the left side of the mandible were

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## LEGENDS FOR FIGURES

Fig. 1. 8 mm craniotomy defect prepared in dried rat skull.

Fig. 2. 15 mm craniotomy defect in dried rabbit skull.

Fig. 3. Dried dog mandible showing fixation of a contemplated 35 mm defect with an ASIF reconstruction plate.

Fig. 4. Dried cynomolgus monkey mandible showing size of a 20 mm discontinuity defect.

Fig. 5. Dried cynomolgus monkey skull showing area available for the creation of experimental defects (8 mm, 15 mm, and 20 mm defects depicted).



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Commercial materials and equipment are identified in this report to specify the investigative procedure. Such identification does not imply recommendation or endorsement or that the material and equipment are necessarily the best available for the purpose. Furthermore, the opinions expressed herein are those of the authors and are not to be construed as those of the U.S. Army Medical Department.

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